

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

IN RE APPLICATION	ATTORNEY DKT NO: 50531
OF: HAUER ET AL.	CONFIRMATION NO.: 6324
SERIAL NO. 10/031,241	GROUP ART UNIT: 1652
FILED: JANUARY 17, 2002	EXAMINER: PAK, YONG D
TITLE: ELECTRON DONOR SYSTEM FOR ENZYMES AND ITS USE IN THE BIOCHEMICAL CONVERSION OF SUBSTRATES	

AMENDMENT AND REPLY UNDER 37 C.F.R. §41.37

Sir,

This Appeal Brief is submitted in response to the Office action of September 26, 2006.

REAL PARTY IN INTEREST

The real party in interest is BASF Aktiengesellschaft, of Ludwigshafen, Germany Reel/Frame 012696/0020 recorded on January 17, 2002.

RELATED APPEALS AND INTERFERENCES

To the best of the undersigned's knowledge there are no related appeals and interferences.

STATUS OF CLAIMS

Claims 1-22 are pending in the application. Claims 1-10, 13-15 and 19-22 are withdrawn from consideration. Claims 11, 12, 16-18 stand rejected and are the subject of this appeal. Additionally claim 12 had been amended and new claims 23 and 24 have been added by Appellants with an amendment filed on April 26, 2007.

STATUS OF AMENDMENTS

An amendment was filed with the filing of this brief on April 26, 2007 to amend claim 12 and add new claims 23 and 24. There were no further amendments to claims subsequent to final action.

SUMMARY OF CLAIMED SUBJECT MATTER

Claim 11 is the sole independent claim involved in this appeal. According to Claim 11, the present claimed invention relates to a method for the enzymatic production of terminally or subterminally hydroxylated fatty acids according to claim 11. *See Application*, page 2, lines 23-33. The method involves the conversion of terminally saturated branched or unbranched fatty acids with 8 to 30 carbon atoms or fatty acid derivatives thereof, selected from C₁-C₄ alkyl esters, amides and anhydrides and isolating the hydroxylated product. *See Application*, page 4, line 41 to page 5, line 9. The conversion takes place in the presence of oxygen and an electron donor system using a cytochrome P450-containing monooxygenase wherein the electron donor system includes an inorganic, non-electrode bound source of electrons and a mediator which is able to transfer electrons from the source of electrons to the enzyme, ie. The cytochrome P450-

containing monooxygenase. *See* Application, page 2, line 35 to page 4, line 6. The non-electrode bound source of electrons is a metal powder which has a lower standard normal potential than the mediator. *See* Application, page 3, lines 4-9, page 9, line 25 to page 27; page 3, lines 11-29.

In other embodiments, the claimed invention can involve hydroxylatable fatty acid is selected from terminally saturated, branched or unbranched C₁₂-C₃₀ fatty acids. *See* claim 12, Application, page 4, line 41-46.

In further embodiments, the enzyme can be a cytochrome P450 mono-oxygenase selected from a) the wild-type enzyme which can be isolated from *Bacillus megaterium* (DSM 32T) or b) a mutant, which can be obtained by amino acid substitution in at least one of positions 26, 47, 72, 74, 87, 188 and 354, of the wild type enzyme (SEQ ID NO: 35). *See* claim,13, Application, page 5, lines 14-19, 41-43.

Furthermore, the electron donor system can be a Zinc/Co(III) sepulchrate. *See* Application, page 3, lines 31-35.

Appellants have also added new dependent claims 23 and 24. Claim 23 recites that the cytochrome P450 mono-oxygenase can be from the CYP102 family. Claim 24 recites that the cytochrome P450 mono-oxygenase can be wild type enzyme from *Bacillus megaterium* or a mutant thereof. *See* Application, page 5, lines 11-43.

GROUND OF REJECTION TO BE REVIEWED ON APPEAL

Whether subject matter of claim 12 is indefinite under 35 USC §112, second paragraph.

Whether subject matter claims 11-12 and 16-18 comply with the written description requirement of 35 USC §112, first paragraph.

Whether subject matter claims 11-12 and 16-18 are enabling under 35 USC §112, first paragraph.

Whether subject matter claims 11-12 and 16-18 are obvious under 35 U.S.C. 103(a) over **Estabrook et al.**, “Application of Electrochemistry for P450-Catalyzed Reaction”, Methods in Enzymology Vol. 272 (1996) pgs, 46-51, (hereinafter “**Estabrook**”) in view of **Creaser et al.**,

“Sepulchrates: a macrobicyclic nitrogen cage for metals ion”, J. Am. Chem. Soc. 1977; 99(9); 3181-3182 (hereinafter “**Creaser**”).

ARGUMENTThe Rejection of Claim 12 35 U.S.C. §112, Second paragraph

The Examiner rejected claim 12 under 35 USC §112, 2nd paragraph as being indefinite asserting that the recitation. Appellants have amended claim 12 in the amendment filed on April 26, 2007 to obviate the above mentioned rejection.

The Rejection of Claims 11-12 and 16-18 under 35 U.S.C. §112, first paragraph

It is the Examiner's position that the subject matter of claims 11-12 and 16-18 are not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventors, at the time the application was filed, had possession of the claimed invention.

The Examiner asserts that there is insufficient descriptive support for the genus comprising any or all cytochrome P450 monooxygenases and genus comprising any or all electron donor system comprising a non-electrode-bound source of electrons. The Examiner further asserts that the specification only teaches a method of hydroxylating fatty acids described in Examples 2-4 of the specification using specific cytochrome P450 monooxygenase obtained from *Bacillus megaterium* and Zinc/Co(III) sepulchrates electron donor system.

The Examiner concludes that these examples are not enough and do not constitute a representative number of all the species to describe the method according to the instant claims. The Examiner also argues that there is no evidence on record to show the relationship between the structure of a *Bacillus megaterium* cytochrome P450 monooxygenase and the structure of a polynucleotide encoding any or all recombinants, variants and mutants of any cytochrome P450 monooxygenase.

The Examiner cites *University of California v. Eli Lilly and Co.* as a basis for the rejections, reciting "a written description of an invention involving a chemical genus, like the description of a chemical species, 'requires a precise definition, such as the structure, formula [or] chemical name' of the claimed subject matter sufficient to distinguish it from other materials." See *University of California v. Eli Lilly and Co.*, 43 USPQ2d 1398 (Fed. Cir. 1997), citing *Fiers v. Revel*, 25 USPQ2d 1601 (Fed. Cir. 1993).

However Appellants respectfully assert that given the state of the art regarding P450 enzymes as well as the significant amount of information provided in the Application regarding the claimed invention, that one of ordinary skill in the art would understand Appellants had possession at the time of filing.

Appellants respectfully submit that since *Lilly*, the Federal Circuit has further clarified that there are many factors which may contribute satisfying the written description requirement. According to *Capon*, the determination of what is needed to support generic claims to biological subject matter depends on a variety of factors, such as (1) the existing knowledge in the particular field, (2) the extent and content of the prior art, (3) the maturity of the science or technology, (4) the predictability of the aspect at issue, and (5) other considerations appropriate to the subject matter. *Capon v. Eshar*, 418 F.3d 1349, 1358, 76 USPQ.2d 1078 (Fed. Cir. Aug 12, 2005).

For example in *Lilly*, the written description requirement was not met because no sequence information indicating which nucleotides constituted the human cDNA at issue was provided in the Application. *See Lilly*, 119 F.3d at 1567. However, in *Capon*, the nucleotide sequence of the chimeric DNA at issue was not required for the written description requirement because “a person experienced in the field of this invention would know that these known DNA segments would retain their DNA sequences when linked by known methods.” *See Capon*, 418 F.3d at 1358. As can be seen, the state of the art of the technology is taken into account when determinations are made with respect to the written description requirement.

This is because “as each field evolves, the balance also evolves between what is known and what is added by each inventive contribution.” *See Id.* *Lilly* involved the discovery of an unknown gene function or structure and so more was required regarding the description of the gene at issue. *See Id.* Appellants respectfully assert that this is far different from the present case.

The instant claims are not directed to an unknown gene function or structure as in *Lilly* but are directed to a specific and superior method for the hydroxylation of fatty acids in the presence of a cytochrome P450 containing mono-oxygenase. Appellants respectfully assert that oxidation reactions using P450 enzymes are well known in the art. *See Estabrook*, pg. 44. The reference **Estabrook**, as cited by the Examiner, discusses oxidation reactions using P450

enzymes. It is further indicated that P450's are "excellent candidates as catalysts" for the synthesizing certain chemicals in such oxidation reactions. See **Estabrook**, pg. 44.

Furthermore, with respect to cytochrome P450 monooxygenases themselves, Appellants respectfully assert that such are well documented and widely studied enzymes.

For example, a search of the National Center for Biotechnology Information¹ provided 45,000+ articles for the search term "cytochromed P450 monooxygenase" up to the filing date of the instant application. Additionally, as indicated in WO 97/49832, all cytochrome P450s are heme binding proteins, associate with specific electron transfer components, and bind molecular oxygen. See WO 97/49832 (filed with IDS), pg. 1, lines 10-31, *background of the invention*. Therefore, there is sufficient structure and function to define what is meant by a recitation of "P450" enzymes. Furthermore, more than 200 cytochrome P450 genes have been described and classified into 36 families, and within a single family, cytochrome P450 proteins have greater than 40% amino acid identity, and greater than 55% within a subfamily. See WO 97/49832 (filed with IDS), pg. 1, lines 10-31, *background of the invention*. Thus, P450 enzymes are a superfamily of related enzymes used for certain oxidation reactions. See *Falkner et al.* "Electrocatalytically driven ω -hydroxylation of fatty acids using cytochrome P450 4A1" Proc. Natl. Acad. Sci. Vol. 92 (1995), pgs. 7705-7709 (presented in IDS).

The above indicates that the state of the art is well studied, and one of ordinary skill in the art would understand what enzymes are meant by cytochrome P450 monooxygenases, and would understand their function in the context of oxidation reactions as claimed. One of ordinary skill in the art would find even further information regarding cytochrome P450 monooxygenases in the instant application. It is indicated on page 1 of the application, that all P450 enzymes have the common function of transferring oxygen atoms to unactivated aliphatic or aromatic X-H (X= -C, -N, -S) bonds. See Application, page 1, lines 21-23. Furthermore, P450 enzymes are further discussed on pages 9-12 of the application, also with selected families shown in table 1, on page 10 of the application.

Additionally, Appellants have provided in the application a detailed method for isolating and producing one particular cytochrome P450 monooxygenase, BM-3 from *Bacillus megaterium*. See Application, pg. 5, lines 11-27, pgs. 14-25. The Examiner asserts that "the examples are not

¹ <http://www.ncbi.nlm.nih.gov/>

enough and does not constitute a representative number of all species to describe a method of hydroxylating the recite fatty acids. *See* Office action, September 26, 2006, pg 4. However, Appellants respectfully assert that in light of the great amount of knowledge and study as outlined above regarding the P450 systems, and that no further examples are necessary, and in fact are more than enough to satisfy the written description requirement.

This is very much unlike *Lilly* wherein there was no information in the patent regarding the genetic cDNA which was subject of the claims. In the case at hand, in light of the state of the art and the information provided in the application, one of skill in the art would understand what is meant by the recitation cytochrome P450 monooxygenases according to the method of the instant claims and recognize that Appellants had possession of the invention at the time of filing.

Appellants further submit that with respect to new claims 23 and 24, Appellants respectfully submit that such claims clearly satisfy the written description requirement. Claim 22 is directed to BM-3 as well as particular mutants as described in the application on page 5, lines 11-39. Furthermore, claim 23 is directed to CYP102 family which is noted in table 1, on page 10 of the application as being involved in reactions for terminal and/or subterminal oxidation of fatty acids. Therefore, Appellants respectfully assert that the subject matter of such claims are clearly described in the application to satisfy 35 USC §112, first paragraph.

The Rejection of Claims 11-12 and 16-18 under 35 U.S.C. §112, first paragraph

The Examiner asserts that while the specification is enabling for a method of hydroxylating fatty acids as described in Examples 2-4 of the specification using a cytochrome P450 monooxygenase obtained from *Bacillus megaterium* and Zinc/Co(III) sephulchrute, the application does not reasonably provide enablement for such a method of hydroxylating fatty acids using any or all variants, mutants and recombinants of any or all cytochrome P450 monooxygenase and any or all electron donor systems comprising any or all electrode-bound source electrons.

The Examiner contends that in view of the great breadth of the claim, it would require an undue amount of experimentation to identify and make the necessary cytochrome

monooxygenase, and to hydroxylate fatty acids. *See* Office action, September 26, 2006, page 10-11. Appellants respectfully traverse the Examiner's assertions.

Enablement under 35 USC §112 requires that the specification describe the invention in such terms that one skilled in the art can make and use the claimed invention. The test is whether a person skilled in the art can make and use the invention without undue experimentation. *See* MPEP §2164.01. It is not necessary that the specification describes in such detail that it enables one skilled in the art to make and use a perfected, commercially viable embodiment of the invention. *See* MPEP §2164.

Some of the factors for consideration in an enablement analysis include : a) the breadth of claims b) the nature of the invention c) the state of the prior art d) the level of one of ordinary skill e) the level of predictability in the art f) the amount of direction provided by the inventor; g) the existence of working examples and h) the quantity of experimentation needed to make or use the invention based on the content of the disclosure. *See In re Wands*, 858 F.2d 731, 737, 8 USPQ2d 1400, 1404 (Fed. Cir. 1988).

Appellants respectfully assert that in view of the state of the art as well as the examples and significant direction provided in the Application, one of ordinary skill in the art could make and use the claimed invention without undue experimentation.

Appellants respectfully submit that little guidance would be needed to conduct the claimed invention. Hydroxylation techniques are well known to the skilled artisan. At the time of the invention, the creation of fatty acids, in generally, was routine.

Furthermore, Appellants respectfully re-assert the remarks made above with respect to the state of the art. As cytochrome P450 monooxygenase are well studied, one of ordinary skill in the art would be very familiar with such enzymes. Furthermore the Application describes how to make and use at least one such enzyme, BM-3 *Bacillus megaterium* as shown in the Examples of the Application. In light of the state of the art and the examples in the Application, one of ordinary skill in the art would be able to conduct the claimed method for hydroxylating fatty acids without undue experimentation. Therefore, Appellants respectfully request the above mentioned rejection be withdrawn.

The Rejection of Claims 11-12 and 16-18 under 35 U.S.C. §103

It is the Examiner's position that the instant claims are obvious over **Estabrook** in view of Creaser. The Examiner asserts that **Estabrook** discloses a method for the enzymatic production of terminally or subterminally hydroxylated fatty acids comprising hydroxylating fatty acids in the presence of an electron donor system, a cytochrome P450 monooxygenase, oxygen, chloride ions and a hydrogen peroxide cleaving enzyme. *See Office action*, September 26, 2006, pg. 15. The Examiner contends that the difference between the reference of **Estabrook** and the claimed invention is that **Estabrook** does not teach a method of producing terminally or subterminally hydroxylated fatty acids using a Zn metal powder form.

The Examiner further asserts that **Creaser** discloses a Zn/Co(III)sepulchrate electron donor system, and teaches that Zn dust causes reduction of the Co(III)sepulchrate mediator within seconds.

In light of this, the Examiner concludes that it would have been obvious to one of ordinary skill in the art to use either Zn dust as originally taught by **Creaser** or Pt as taught by **Estabrook** in hydroxylating fatty acids using a metal/Co(III)sepulchrate electron donor system.

According to §103, in order to establish a prima facie case of obviousness, there must be (1) some suggestion or motivation to modify the references, (2) reasonable expectation of success and (3) the prior art reference must teach or suggest all of the claim limitations. *See In re Vaeck*, 947 F.2d 488, 20 USPQ2d 1438, MPEP §2143. Appellants respectfully submit that in the present case, there is no suggestion or motivation to modify the references as indicated by the Examiner.

Appellants respectfully assert that the Examiner is engaged in impermissible hindsight reasoning to modify the references using the present application as a blueprint. *See* MPEP 2145.X.A.

Appellants respectfully submit that the mere fact that a reference can be combined or modified does not render the resultant modification obvious unless the prior art also suggests the desirability of the combination. *See In re Mills*, 916 F.2d 680, 682, 16 USPQ.2d 1430; MPEP 2143.01.III.

Appellants respectfully assert that there is not the slightest piece of guidance in **Creaser** to use Zn dust in a biochemical system. There is no experimental guidance, nor is there even

hypothetical guidance in the cited reference. To the contrary, at the end of the reference, it is speculated to use in organic and inorganic chemistry to use as therapeutic agents – still without any experimental guidance. See **Creaser**, pg. 3182, right col., 2nd ¶. Potential biochemical applications are not at all mentioned.

Appellants also respectfully note that there is not the slightest suggestion or piece of guidance in **Estabrook** to replace the highly sophisticated electrode system by a metal powder. Furthermore, although citing the **Creaser** in a footnote (FN 6), the **Estabrook** reference says nothing of using Zn powder and instead uses a Pt electrode with the Co(III)sepulchrate as a mediator.

Furthermore, looking at the state of the art in general, and not only at the cited references, Appellants respectfully assert that there is still no motivation for use of Zn powder for hydroxylation of fatty acids. Appellants wish to note the work of *Fang*, which also references the work of *Faulkner*, both of which were noted in Appellants IDS. See *Fang*, “Dithionite-Supported Hydroxylation of Palmitic Acid by Cytochrome P450BM-3” *Drug Metabolism and Disposition* Vol. 24, No. 11 (1996) pgs, 1282-1285; *Faulkner et al.* “Electrocatalytically driven ω -hydroxylation of fatty acids using cytochrome P450 4A1” *Proc. Natl. Acad. Sci.* Vol. 92 (1995) pgs 7705-7709.

Starting from the electrode-based electron donor system of *Faulkner*, *Fang* suggests employing dithionite as a reducing agent for the P450 enzyme. Thus, one of ordinary skill in the art, starting from the electrode-based electron donor system of **Estabrook**, would have expected success with a reducing agent which is soluble in the reaction mixture, as for example dithionite, rather than the metal-based system of *Creaser* and *Sargeson* (US 4,497,737; cited by Examiner) which according to *Sargeson* (See col. 4, lines 60-65) was regarded as a suitable redox system for inorganic and organic synthesis rather than biosynthesis. Thus, in view of the state of the art, one of ordinary skill in the art would find no motivation to use a metal in powder form as a source of electrons.

Appellants respectfully assert that the claimed invention also produces superior and unexpected results. Unexpected properties can be used to show that a claimed invention is not obvious over the cited references. See *Dillon*, 919 F.2d 688, 692, 16 USPQ2d 1897 (Fed. Cir. 1990); MPEP 2144.08.II.B. Appellants refer to data from experimental work submitted in the


reply of February 12, 2004. Similar to the experiments performed by **Estabrook**, and summarized in Table 1 of the reference, the Appellants compared reaction rates for the BM-3 mutant F87-A under conditions similar to the cited art and also to the claimed invention. Artificial substrate 12-pNCA was used as the enzyme substrate to measure activity via an optical test. Enzyme activity was measured in separate reactions in either the presence of NADPH or electrolysis/Pt-electrode (Estabrook) and NADPH or Zn dust and Co(III)sepulchrates as electron sources. With the NADPH reactions set to 100%, the results showed that Zn dust and Co(III)sepulchrates reaction had a relative reaction almost twice as great as that of the **Estabrook** example. These data show that the electron donor system of the instant claims is more suitable for enzyme reactions as claimed because of the higher reaction rates. Such results cannot be expected from the disclosures of the cited references and the knowledge in the art at the time of filing.

In view of the foregoing, Appellants respectfully assert that no prima facie case of obviousness can be established, and accordingly, request the 35 USC §103 rejection be withdrawn.

CONCLUSION

In view of the foregoing, Appellant reasserts and maintains its position set forth in its Appeal Brief. Please charge any shortage in fees due in connection with the filing of this paper, including extensions of time, to Deposit Account No. 14-1437

Respectfully submitted,
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CLAIMS APPENDIX

The Attached APPENDIX is a correct listing of the current pending claims

1. An electron donor system for transferring electrons to enzymes with redox properties, wherein the system comprises an inorganic, non-electrode-bound source of electrons and a mediator which is able to transfer electrons from the source of electrons to the enzyme.
2. An electron donor system as claimed in claim 1, wherein the enzyme is a cytochrome P450-containing enzyme.
3. An electron donor system as claimed in claim 2, wherein the enzyme is a mono oxygenase (E.C. 1.14).
4. An electron donor system as claimed in claim 1, wherein the mediator has a standard normal potential in the region of less than about -0.4 V.
5. An electron donor system as claimed in any of the preceding claims, wherein the mediator is selected from cobalt(III) sepulchrates, methylviologen, neutral red, riboflavin, ruthenium triacetate, FMN and FAD.
6. An electron donor system as claimed in claim 1, wherein the source of electrons is a metal with a lower standard normal potential than the mediator.
7. An electron donor system as claimed in claim 6, wherein the source of electrons is metallic zinc.
8. An electron donor system as claimed in claim 1, selected from the systems:
 - Zn/cobalt(III) sepulchrates and
 - Zn/neutral red.

9. A method for the enzymatic transfer of oxygen to a hydrocarbon-containing hydrogen donor molecule, which comprises incubating the hydrogen donor molecule in a reaction medium comprising the oxygen-transferring enzyme and an electron donor system as claimed in claim 1 in the presence of oxygen under reaction conditions.
10. A method as claimed in claim 9, wherein the hydrogen donor molecule is selected from compounds of the formula.
11. A method for the enzymatic production of terminally or subterminally hydroxylated fatty acids, which comprises
 - a) converting a fatty acid selected from terminally saturated, branched or unbranched fatty acids with 8 to 30 carbon atoms or fatty acid derivative thereof, selected from C₁-C₄ alkyl esters, amides and anhydrides, in the presence of an electron donor system using a cytochrome P450 monooxygenase and oxygen wherein said electron donor system comprises an inorganic, non-electrode bound source of electrons and a mediator which is able to transfer electrons from the source of electrons to the enzyme, wherein said enzyme is a cytochrome P450-containing monooxygenase (E.C. 1.14), and wherein the source of electrons is a metal in powder form with a lower standard normal potential than the mediator; and
 - b) isolating the hydroxylated product(s).
12. A method as claimed in claim 11, wherein the hydroxylatable fatty acid is selected from terminally saturated, branched or unbranched C₁₂-C₃₀ fatty acids.
13. A method as claimed in claim 9, wherein the enzyme is a cytochrome P450 monooxygenase selected from:
 - a) the wild-type enzyme which can be isolated from *Bacillus megaterium* (DSM 32T); or

- b) a mutant, which can be obtained by amino acid substitution in at least one of positions 26, 47, 72, 74, 87, 188 and 354, of the wild-type enzyme (SEQ ID NO: 35).
- 14. A method as claimed in claim 13, wherein a single mutant selected from F87A, F87V, L188K, V26T, R47F and V26T.
 - 15. A method as claimed in claim 13, wherein the mutant has in position 87 the mutation F87A or F87V and at least one other of the following mutations: L188K, A74G, R47F and V26T.
 - 16. A method as claimed in claim 11, wherein the electron donor system is zinc/Co(III) sepulchrate.
 - 17. A method as claimed in claim 11, wherein at least stage a) is carried out in the presence of chloride ions.
 - 18. A method as claimed in claim 11, wherein at least stage a) is carried out in the presence of a hydrogen peroxide-cleaving enzyme.
 - 19. A bioreactor for use for producing ω -hydroxylated fatty acids, which comprises immobilized monooxygenase and an electron donor system as claimed in claim 1 in a liquid reaction medium.
 - 20. A detection method for fatty acid monooxygenases, which comprises
 - a) incubating an analyte suspected of having enzymic activity with an ω -hydroxylatable fatty acid or fatty acid derivative which has a terminal chromophore or fluorophore which can be eliminated, in the presence of an electron donor system as claimed in claim 1; and

- b) determining the elimination of the chromophore or fluorophore qualitatively or quantitatively.
21. A method as claimed in claim 20, wherein the conversion is carried out in the presence of a hydrogen peroxide-cleaving enzyme and, where appropriate, in the presence of chloride ions.
22. A test kit comprising an electron donor system as claimed in claim 1.
23. A method as in claim 11, where the cytochrome P450 mono-oxygenase is from the CYP102 family.
24. A method as in claim 11, wherein the cytochrome P450 mono-oxygenase is selected from the group consisting of:
- a) the wild-type enzyme which can be isolated from *Bacillus megaterium* (DSM 32T); or
 - b) a mutant, which can be obtained by amino acid substitution in at least one of positions 26, 47, 72, 74, 87, 188, 354, of the wild-type enzyme (SEQ ID NO:35).

EVIDENCE APPENDIX

This Evidence Appendix includes the following:

	P450 enzymes	electron source	mediator	substrate	reaction rate [eq/min]*	relative rate
Estabrook et al.	BM-3	NADPH	-	lauric acid	900	100 %
Estabrook et al.	BM-3	electrolysis/Pt- electrode	Co(II)sep	lauric acid	110	12 %
Inventor	BM-3 F87A	NADPH	-	12-pNCA	574 ± 28	100 %
Inventor	BM-3 F87A	zinc dust	Co(II)sep	12-pNCA	125 ± 6	22 %

RELATED PROCEEDINGS APPENDIX

None